



First Self-Replicating Synthetic Bacterial Cell

J. CRAIG VENTER INSTITUTE

Synthetic Genomics Research and the First Self-Replicating Synthetic Bacterial Cell Constructed by Scientists at the J. Craig Venter Institute

Frequently Asked Questions (FAQ) ►

Q: Is your work in creating a synthetic bacterial cell “creating life from scratch”?

A: No we do not consider this to be “creating life from scratch” but rather we are creating new life out of already existing life using synthetic DNA to reprogram the cells to form new cells that are specified by the synthetic DNA.

Q: Why construct a synthetic cell?

A: We believe that the ability to “write the genetic code” as we describe synthetic genomics research will enable a better understanding of the fundamentals of living cells. It will also enable us to direct cells and organisms to perform jobs, such as creating clean water or new biofuels that natural species cannot currently do to the needed scale and efficiencies.

Q: How is this different than standard molecular biology/genetic engineering, etc?

A: Scientists have long been able to change and/or modify single genes or small sets of genes. Most genetic alterations that people know about today are through engineering of crops, which involves adding or altering less than 10 genes out of the tens of thousands that are contained in most organisms or plants. Synthetic genomics is different in that scientists start with digital information in the computer, which allows for the design of entire synthetic chromosomes to replace existing chromosomes in cells. The first self-replicating synthetic bacteria cell constructed by scientists at the JCVI has more than 1 million base pairs of DNA, almost 1,000 genes, and involved the complete replacement of genetic material in the cell.

Q: Describe the potential applications of a synthetic cell. What is the impact of this area of science and the resulting technologies?

A: The work to create a synthetic cell will have a profound and positive impact on society in that it will enable a better understanding of the fundamentals of biology and how life works. It will lead to new techniques and tools for advanced vaccine and pharmaceutical development, and will continue to enable the development of new biofuels and biochemicals. As well these technologies could be used to create clean water, new sources of food, textiles, bioremediation, etc. Dr. Venter and the teams at JCVI and the company Synthetic Genomics Inc (SGI) believe that this science has the potential to be a major wealth driver for societies. A recent report, "Synthetic Biology: scope, applications and implications," from the Royal Academy of Engineering in the UK states, "Synthetic biology has the potential to create another raft of major new industries, the development of which is likely to have profound implications for the future of the UK, European and world economies." http://www.raeng.org.uk/news/publications/list/reports/Synthetic_biology.pdf

Q: How many researchers were involved in constructing the first self-replicating, synthetic bacterial cell?

A: For nearly 15 years researchers at the JCVI have been working on various aspects of the synthetic genomics research and the quest to create the first synthetic cell. In the early stages there were just a few researchers on the team but it has grown over the years to include approximately 24. They are: J. Craig Venter, Hamilton Smith, Clyde Hutchison, John Glass, Dan Gibson, Carole Lartigue, Gwyn Benders, Vladimir N. Noskov, Ray-Yuan Chuang, Mikkel A. Algire, Michael G. Montague, Li Ma, Monzia M. Moodie, Chuck Merryman, Sanjay Vashee, Radha Krishnakumar, Nacyra Assad-Garcia, Cynthia Andrews-Pfannkoch, Evgeniya A. Denisova, Lei Young, Zhi-Qing Qi, Thomas H. Segall-Shapiro, Christopher H. Calvey, and Prashanth P. Parmar.

Q: What are the risks associated with synthetic organisms? Do the risks of these technologies overshadow the potential benefits?

A: As with any new area of science, medicine or technology, synthetic genomics has the potential to be used for great societal benefit (biofuels, vaccines and pharmaceuticals, clean water, bioremediation, etc), but it could also be used for negative purposes. So called dual use technologies need to be carefully discussed and reviewed both at the government level (Federal, state and local) both in the US and globally, as well as in accessible forums for bioethicists, educators, students, media and laypeople to learn about the science and understand these risks and benefits.

Dr. Venter and his teams at both the JCVI and at SGI, have, as the leaders of this field been driving these ethical and societal implications since the beginning of the research (for nearly 15 years). The policy team at JCVI has been funded to conduct an already completed study on the options for governance of this field as well as an ongoing study of the societal issues this work raises. Many other countries are reviewing and discussing this area of science and as such numerous reports and reviews have also been conducted. Please see the JCVI Fact Sheet on Ethical Considerations and Societal Implications for more detail on this work.

Q: Does this work have anything to do with humans/human research?

A: No. All synthetic genomics work to date, both at the JCVI and elsewhere has focused on microorganisms. It is anticipated that given how little is known about human biology that no applications of this work will or should be attempted in humans. The way that this research will impact human lives is through the numerous applications such as new vaccines, pharmaceuticals, biofuels, etc.

Q: What safeguards/controls are in place to protect against accidental environmental release?

A: This is an extremely important question for this research and as such has been a big focus for the researchers at JCVI, as well as the company Synthetic Genomics Inc. Building on the longstanding and successful history in molecular biology of millions of experiments engineering and using organisms like *E. coli* to conduct research, JCVI and Synthetic Genomics researchers will be able to engineer synthetic bacterial cells so they cannot live outside of the lab or other production environments. This is done by, for example, ensuring that these organisms have built in dependencies for certain nutrients without which they cannot survive. They can also be engineered with so called "suicide genes" that kick in to prevent the organism from living outside of the lab or environment in which they were grown.

Q: Are the bacteria used in the proof of principle work in creating the first synthetic cells (*Mycoplasma genitalium*, *Mycoplasma mycoides*, *Mycoplasma capricolum*) dangerous or pathogenic to humans or animals?

A: There are many species and subspecies of *Mycoplasmas*, many with similar sounding names. However, the *Mycoplasmas* the JCVI team is working with—wild type *M. mycoides* subspecies *capri* and *Mycoplasma capricolum* subspecies *capricolum* are not dangerous to humans. However, both are commonly found in goat herds worldwide, and are often isolated from goats in the US. The organisms typically cause mild mastitis and polyarthritis in some goats. Importantly, it is not the agent of contagious caprine pleuropneumonia (CCPP). Rather, that is the capripneumoniae subspecies of *Mycoplasma capricolum*. It is also not the same as *M. mycoides* subspecies *mycoides*, which is the agent of contagious bovine pleuropneumoniae (CBPP or lung plague in cattle). While the *M. mycoides mycoides* and *M. capricolum capripneumoniae* subspecies are endemic in Africa and a huge burden for cattle and goat populations there, the organisms are not present in the Western Hemisphere. In the US research on those pathogens can be done at the U.S. Department of Agriculture's Plum Island Animal Disease Center. The synthetic *M. mycoides* organism has 14 genes deleted or inactivated, including genes that are predicted to be critical to virulence. While it might be capable of infecting goats, and could potentially cause disease, this has not been tested.

Mycoplasma genitalium, the species whose genome we previously synthesized is a sexually transmitted parasite of the human urogenital tract that causes urethritis and is implicated in adverse pregnancy outcomes. Perhaps as much as 1% of adults may be infected, often asymptotically. The JCVI team is still working to boot up this first synthetic bacterial genome. The extremely slow growth rate of this organism makes experimentation very time consuming (this organism's slow growth also compelled us to resort to working with the faster growing species *M. mycoides* that we report on now). The JCVI efforts to boot up *M. genitalium* synthetic cell have yielded new understandings about the peculiar biology of this simple cell that we believe will soon result in our booting up this synthetic genome as well.

Q: What are the next steps for this research at JCVI?

A: The work to create the first self-replicating, synthetic bacterial cell was an important proof of concept. The team at JCVI has learned a lot from the nearly 15 years it has taken to get to this successful stage. From this proof of concept experiment the team is now ready to build more complex organisms with useful properties. For example, many, including scientists at SGI, are already using available sequencing information to engineer cells that can produce energy, pharmaceuticals, and industrial compounds, and sequester carbon dioxide. The team at JCVI is already working on their ultimate objective, which has been to synthesize a minimal cell that has only the machinery necessary for independent life. Now that a cell can be synthesized from a synthetic genome in a simple near-minimal bacterial cell, it becomes possible for the team to test for the functionality of a genome. They can whittle away non-essential DNA regions from the synthetic genome and repeat transplantation experiments until no more genes can be disrupted and the genome is as small as possible. This minimal bacteria cell will enable a greater understanding of the function of every gene in a cell and a new vision of cells as understandable machines comprised of biological parts of known function.

Q: Is this research patented?

A: Over the course of the 15 years it has taken to construct the first self-replicating synthetic bacterial cell, the team at JCVI has had to develop new tools and technologies to enable this feat. SGI has funded the work at JCVI in exchange for exclusive intellectual property rights. SGI has filed 13 patent family applications on the unique inventions of the JCVI team. SGI believes that intellectual property is important in the synthetic genomics/biology space as it is one of the best means to ensure that this important area of basic science research will be translated into key commercial products and services for the benefit of society. SGI intends to provide licenses to its synthetic genomics patents.

Q: Has there been any review of this work by the US government? Review by any other organizations?

A: The synthetic genomics research at JCVI has undergone review at the highest levels of the US government. Beginning in 2003 with the publication of the research at JCVI in creating the synthetic phiX174 ("Generating a synthetic genome by whole genome assembly: phiX174 bacteriophage from synthetic oligonucleotides." Smith et al, PNAS 2003 Dec 23;100(26):15440-5. Epub 2003 Dec 2.), and including the most recent research and publication on creating the first self-replicating synthetic bacterial cell, the work has been reviewed by White House offices including the Office of Homeland Security and Office of Science and Technology Policy, the National Science Advisory Board for Biosecurity (NSABB), the

Department of Energy, the National Institutes of Health, and others. As well the work has been reviewed by independent bioethics groups since 1997. Senior US government officials including those at the NIH were briefed and allowed to review our study prior to publication.

Q: What, if any, types of legislation or regulation should be applied to this area of research?

A: We think that it is prudent, as is being proposed by HHS, to require DNA synthesis companies to screen synthesis requests against harmful agents. In 2004, the JCVI's Policy team, along with the Center for Strategic & International Studies (CSIS) and the Massachusetts Institute of Technology (MIT) were funded by the Alfred P. Sloan Foundation to conduct a series of workshops and public sessions over a 20-month period to discuss the ethical and societal implications of synthetic genomics. Over the course of the study, the group explored the risks and benefits of the emerging technology, as well as possible safeguards to prevent abuse, such as bioterrorism. In October of 2007 the group published their findings in a report, outlining options for the field and its researchers moving forward.

Most recently in December of 2008, JCVI received funding from the Alfred P. Sloan Foundation to examine ethical and societal concerns that are associated with the developing science of synthetic genomics. The ongoing research is intended to inform the scientific community as well as educate our policymakers and journalists so that they may engage in informed discussions on the topic.

Q: What is the relationship between JCVI and the company Synthetic Genomics Inc and in particular how it relates to the research to create the first synthetic bacterial cell?

A: JCVI is a not-for-profit, genomic-focused basic research organization. Synthetic Genomics Inc (SGI) is a privately held company founded to apply synthetic genomic-driven commercial solutions in a variety of global challenges including energy and the environment. Both organizations were founded by and are headed by Dr. J. Craig Venter. Since 2005, SGI has funded approximately \$30 million of synthetic genomics basic science research at the JCVI. SGI has exclusive assignment of intellectual property rights on this work and has filed approximately 13 patent application families to date.